INVESTIGATOR'S ANNUAL REPORT

National Park Service

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Permit#: SHEN-2004-SCI-0008	
Park-assigned Study Id. #: SHEN-00295	
Project Title: Floral Scents of Hybrids: Bridge or Barrier to Interspecific Gene Flow?	
Permit Start Date: Jun 02, 2004	Permit Expiration Date Sep 01, 2008
Study Start Date: Jun 02, 2004	Study End Date Sep 01, 2008
Study Status: Continuing	
Activity Type: Research	
Subject/Discipline: Ecology (Aquatic, Marine, Terrestrial)	

Objectives:

Recent advances in the application of transgenic crops have heightened awareness of the potential ecological impacts of these emerging technologies. One area of concern is the risk of unintended gene flow from genetically modified crops to unintended target plants or wild relatives. Basic research in the evolutionary processes of speciation and hybridization may have important insights to offer investigators of this phenomenon. The behavior of insect pollinators is among the most significant factors that determine whether two populations or species are reproductively isolated. There is a rich history of research on what factors attract insects to flowers. However, only recently, with the use of powerful genetic and statistical tools, have ecologists and evolutionary biologists been able to accurately track gene flow in populations and accurately measure the contribution of individual plant traits to reproductive success. Integrative research on plant morphology, genetics, and pollinator interactions has now shown how a few plant traits, such as the color and shape of flowers, affect reproductive barriers and gene flow. However, one trait that is extremely important as a pollinator attractant, floral scent, has gone largely unstudied from this perspective.

We propose to use species of Asclepias (milkweeds) to investigate how floral scent is inherited by hybrids and how scents affect gene flow between species. Do the scents of hybrids accelerate or impede the rate of gene flow between species? A wealth of prior research has documented the effects of floral traits on the reproductive success of milkweeds and the roles of pollinators in mediating these effects. Milkweeds provide an illuminating system because of the rarity of successful hybridization, even though many species co-occur over vast areas. Even in the system in which we are working, in which hybridization has been well documented, there are strong barriers to hybridization. It has been hypothesized that rare F1 hybrids have morphological characteristics that bridge reproductive isolation.

We propose to rigorously test the roles of hybrids as bridges promoting gene flow between species, with emphasis on an important, but often overlooked, attribute of floral morphologyfloral scent. Our experiments will provide novel insights into the phenotypic characteristics of hybrids, the underlying genetics of these traits, the effects of these traits on the patterns of mating among hybrids and their parental populations, and the impact of these mating patterns on gene flow between species. Using a combination of observational studies of natural populations, controlled crossing experiments, and controlled pollination experiments, we will integrate mechanistic and realistic explanations for the affect of hybrid scents on gene

flow.

The proposed research will have significant impacts on training, outreach to underrepresented groups, and applied scientific disciplines. Mississippi State University provides an excellent opportunity to increase the research opportunities of African American students. The postdoctoral associate and undergraduate students will attend national meetings to present the results of their contributions to the project. The results of the proposed research will likely impact disciplines beyond ecology and evolutionary biology. Insights from this investigation could make a significant contribution to safely cultivating genetically engineered crops. Also, there are several threatened and endangered species of Asclepias, and this genus is known as a crucial food plant of the Monarch butterfly, which is of considerable conservation concern. A better understanding of the reproductive biology and hybridization dynamics of A. exaltata and A. syriaca may contribute to the conservation of rare milkweeds and the insects that depend on them.

Findings and Status:

In 2004 we collected data on the identities of insect pollinators to Asclepias exaltata, A. syriaca, and their hybrid, the rates of flower visitation by each pollinating species, and the rates of pollen transfer effected by each pollinating species. Pollination rates were measured for both parental plant species and their hybrid. Floral scent samples were collected from both parental species and their hybrids and these samples were analyzed by gas chromatography-mass spectroscopy. Forty-nine plants were observed for 30 min observation periods during which pollinator species were identified and the duration of their foraging visits to flowers were measured. Asclepias syriaca received significantly more visits by pollinators than either Asclepias exaltata or the hybrid; the latter two did not differ in visitation rates. Insect species differed significantly in the rates at which they visited flowers, regardless of plant species. Silver-spotted skippers (a butterfly), bumblebees, small solitary bees, and honeybees were most frequent flower visitors. Plants were also observed to measure the rates of pollen removal and deposition per insect visit. Asclepias exaltata had significantly lower rates of pollen removal than A. syriaca or the hybrid; the latter two did not differ significantly. Insects also differed in rates of pollen removal from the plants. Bumblebees had the highest removal rate on hybrids, whereas honeybees had the highest removal rates on A. syriaca. However, silver-spotted skippers were the only insects observed to successfully remove pollen from both species and their hybrid. These data suggest that the silver-spotted skipper is the most likely agent of interspecific pollen transfer in this hybrid zone. The high rates of insect visitation to A. syriaca, coupled with the low rates of pollen removal from A. exaltata suggest that matings between A. syriaca and hybrids should occur more frequently and matings between A. exaltata and hybrids, suggesting greater potential for introgression of traits from

Floral scents were collected from 14 plants representing both species and their hybrid. Scents were collected from flowers in the field by bagging flowers and creating a continuous airflow over the flowers and through an adsorbent filter using a pump. Collected scents were eluted into a solvent and transported to University of South Carolina for analysis in the lab of Dr. Rob Raguso. Both species and their hybrid differed in floral scent composition. Asclepias syriaca has a scent composed of a large number of compounds at high concentrations and has a noticeably strong, pleasant scent. Asclepias exaltata has few compounds, all at low concentration and has a weak, unpleasant scent. Hybrid plants have strong scents reminiscent of A. syriaca. However, two compounds found only at trace levels in A. syriaca are present at very high levels in the hybrid. This is one of the first documented cases of unique scent profiles documented from the flowers of hybrids relative to their parents. The scent similarity between A. syriaca and hybrid plants reinforces the supposition from insect visitation data that matings between A. syriaca and hybrids are most likely and that introgression of traits into A. syriaca is more likely than into A. exaltata.

Results from 2004 will be bolstered by increase sample sizes in 2005 and additional data will be collected on pollination dynamics in the hybrid zone, as well as the distribution of genetic markers across the hybrid zone.

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For this study, were one or more specimens collected and removed from the park but not destroyed during analyses? Yes		
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Full name of college or university:	Annual funding provided by NPS to university or college this reporting year:	
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